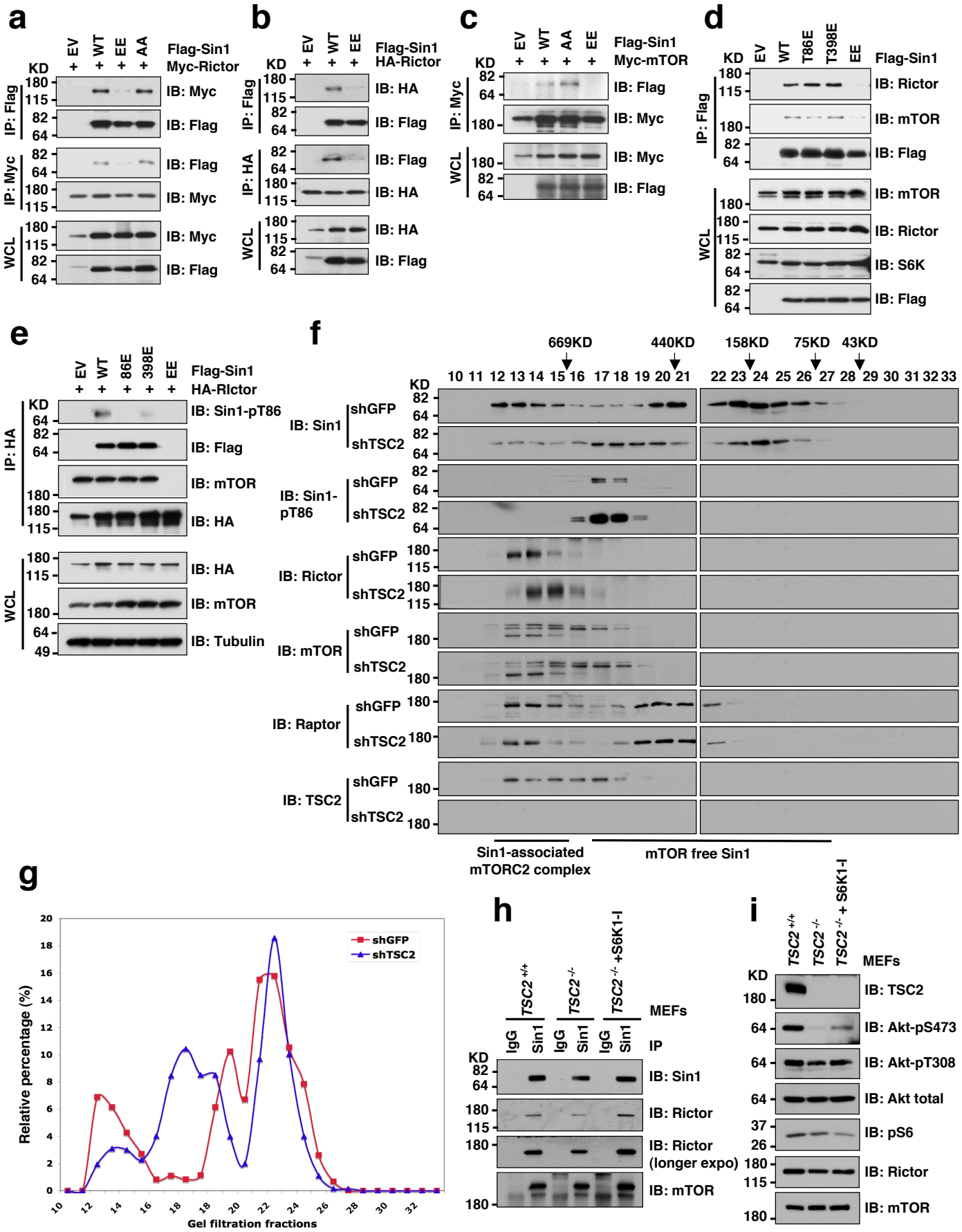
Figure S3


a
EV
WT
EE
AA
Flag-Sin1
Myc-Rictor
KD
IP: Flag
IP: Myc
WCL
IB: Myc
IB: Flag
b
Flag-Sin1
HA-Rictor
IP: HA
IB: HA
c
Flag-Sin1
Myc-mTOR
d
T86E
T398E
IB: Rictor
IB: mTOR
IB: S6K
e
86E
398E
HA-RIctor
IB: Sin1-pT86
IB: Tubulin
f
669KD
440KD
158KD
75KD
43KD
shGFP
shTSC2
IB: Sin1
IB: Sin1-pT86
IB: Rictor
IB: mTOR
IB: Raptor
IB: TSC2
Sin1-associated mTORC2 complex
mTOR free Sin1
g
shGFP
shTSC2
Relative percentage (%)
Gel filtration fractions
h
TSC2+/+
TSC2-/-
TSC2-/- +S6K1-I
MEFs
IgG
Sin1
IP
IB: Sin1
IB: Rictor
IB: Rictor (longer expo)
IB: mTOR
i
TSC2+/+
TSC2-/-
TSC2-/- + S6K1-I
MEFs
IB: TSC2
IB: Akt-pS473
IB: Akt-pT308
IB: Akt total
IB: pS6
IB: Rictor
IB: mTOR